# Correspondence

## Reply

*To the Editor:*

Amoxicillin can be obtained as a pure acid (molecular formula: $C_{16}H_{19}N_3O_5S$, molecular weight: 365.4) or as a trihydrate (molecular formula: $C_{16}H_{19}N_3O_5S \cdot 3H_2O$, molecular weight: 419.45). The intravenous form, previously used in Europe, is typically a sodium salt of the pure acid. The oral form, currently in use worldwide, is the trihydrate. Since the mid 1990s, we have used the sodium salt of the pure acid form of amoxicillin, at a concentration of 0.01 mol, obtained from Sigma-Aldrich Chemicals (St Louis, Mo), for our skin testing[1] The key point is not whether you force amoxicillin, in either form, into solution at a high or low pH, as noted by Tsuji and coworkers,[2] but what is the solubility of amoxicillin at a physiologic pH near 7.4 where you would want to use it for skin testing and what is the pH of the high-concentration amoxicillin skin testing materials used by Montañez and coworkers?[3] The U-shaped solubility for all amino-penicillins in aqueous solution is noted by Tsuji and coworkers in their Figure 5.[2]

The Sigma Chemical package insert for amoxicillin, as the pure acid (Product Number A 8523), states that this product is soluble in 1 mol of ammonium hydroxide up to 50 mg/mL, yielding a clear, colorless to light yellow solution. A study of the solution stability of amoxicillin sodium at 10 mg/mL in sterile water noted a starting pH of 8.7, with a fall from 0.1 to 0.4 pH units over a range of 10% to 80% amoxicillin loss.[4]

In any case, use of amoxicillin, at 0.01 mol or about 3.65 mg/mL in buffered saline at pH 7.4, is adequate to safely evaluate patients for IgE-mediated amoxicillin allergy.[5] Amoxicillin appears to be a valuable adjunct to commercially available penicilloyl-polylysine (Pre-Pen; ALK, Round Rock, Tex), used at $6 \times 10^{-5}$ mol, and native penicillin, like amoxicillin used at 0.01 mol, when evaluating patients with a history of penicillin allergy to identify additional skin-test–positive individuals. This may reduce the number of positive oral challenge reactions, though this has not been proven. All patients with negative penicillin skin test results—whether amoxicillin is used or not—should still undergo an oral amoxicillin challenge.[6]

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### REFERENCES

1. Macy E, Richter PK, Falkoff R, Zeiger RS. Skin testing with penicilloate and penilloate prepared by an improved method: amoxicillin oral challenge in patients with negative skin test responses to penicillin reagents. J Allergy Clin Immunol 1997;100:586-91.
2. Tsuji A, Nakashima E, Hamano S, Tamana T. Physiochemical properties of amphoteric β-lactam antibiotics, I: stability, solubility, and dissolution behavior of amino penicillins as a function of pH. J Pharm Sci 1978;67:1059-66.
3. Montañez MI, Torres MJ, Perez-Inestrosa E, Blanca M. Clarification concerning amoxicillin skin testing. J Allergy Clin Immunol 2011;XXX:XXX-XXX.
4. Concannon J, Lovitt H, Ramage M, Tai LH, McDonald C, Sunderland VB. Stability of aqueous solutions of amoxicillin sodium in the frozen and liquid states. Am J Hosp Pharm 1986;43:3027-30.
5. Lin E, Saxon A, Riedl M. Penicillin allergy: value of including amoxicillin as a determinant in penicillin skin testing. Int Arch Allergy Immunol 2010;152:313-8.
6. Macy E, Goldberg B, Poon KWT. Commercial anti-penicillin-IgE FEIAs are not useful in diagnosing penicillin allergy in patients with remote histories of penicillin allergy. Ann Allergy Asthma Immunol 2010;105:136-41.

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# Correspondence

## Clarification concerning amoxicillin skin testing

*To the Editor:*

Regarding the controversy concerning the solubility of amoxicillin for skin testing,[1,2] an important misunderstanding exists. Macy states that amoxicillin's solubility in water at pH 7 is 4 mg/mL. Thus, skin testing using 20 mg/mL of amoxicillin is an irritant because it requires a nonphysiologic pH to ensure amoxicillin's solubility in solution.[2] Macy does not take into account that three forms of amoxicillin exist, with their corresponding Chemical Abstracts Service (CAS) registry numbers.[3]

The active pharmaceutical principle is the molecule of amoxicillin (CAS 26787-78-0), which provides the antibiotic activity. Based on this, 2 types of amoxicillin are available (Fig 1): the injectable type is a sodium salt (CAS 34642-77-8)[3] that can be easily dissolved at physiologic pH in water. In fact, 200 mg/mL is the concentration of amoxicillin allowed for parenteral use and for which complete solubility is necessary in order to avoid adverse effects. The other type is the trihydrate form (CAS 61336-70-7)[3] used for oral administration. It displays a carboxylic acid functionality and cannot be easily dissolved in water. The *European Pharmacopeia 5.0* states that the water solubility of these 2 types of amoxicillin is different, with amoxicillin trihydrate described as slightly soluble and amoxicillin sodium as very soluble. The less water-soluble form (trihydrate) is the one Macy has used for skin testing, and it is therefore impossible to reach a concentration of 20 mg/mL. Thus, he wrongly concludes that false-positive data have been reported in most studies.[2]

Since the 1980s all these considerations have been taken into account, which is why the injectable form of amoxicillin was selected.[4-6] This has enabled the diagnosis based on skin test in a considerable number of patients who would otherwise have been missed if this procedure had not been carried out.[5,6] Based on this evidence, this practice is now recommended by the European Network of Drug Allergy for skin testing at 20 mg/mL.[6] It is an inexcusable mistake to confuse the 2 structures of amoxicillin even though the difference consists of a single atom, either sodium or hydrogen, that provides the salt and the trihydrate form with their respective properties.

The author has extended the same confusion to ampicillin. Similarly, 3 compounds are registered in CAS,[3] corresponding to ampicillin (CAS 69-53-4), the trihydrate form (CAS 7177-48-2), and the sodium salt (CAS 69-52-3). The oral trihydrate form is slightly soluble in water, and the injectable sodium salt is more soluble (50 mg/mL).

There is an effect of pH on solubility. The ionization constants have been calculated for these 2 aminopenicillins.[7] The $pK_a$'s of amoxicillin are 2.67, 7.11, and 9.55, whereas the $pK_a$'s for ampicillin are 2.67 and 6.95. At a pH equal to the isoelectric point, they exist essentially as zwitterions and, in this form, are more stable and less soluble in water. Certainly, different forms of the same chemical compound can exhibit different physical and chemical properties, including different solubility and dissolution profiles, which, in turn, can affect the bioavailability and stability of the drug.

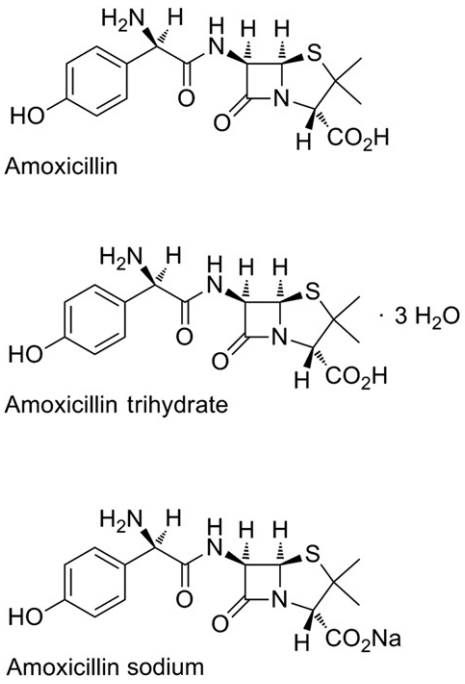


**FIG 1.** Chemical structures of amoxicillin.

With this correspondence we hope to have clarified this important issue.

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### REFERENCES

1. Romano A, Gaeta F, Valluzzi RL, Caruso C, Rumi G, Bousquet PJ. IgE-mediated hypersensitivity to cephalosporins: cross-reactivity and tolerability of penicillins, monobactams, and carbapenems. J Allergy Clin Immunol 2010;126:994-9.
2. Macy E. Penicillin allergy might not be very common in subjects with cephalosporin allergy. J Allergy Clin Immunol 2011;127:1638.
3. Budavari S, O'Neil M, Smith A, Heckelman P, Obenchain J, Gallipeau A, et al. The Merck index, 13th ed. Whitehouse Station (NJ): Merck & Co, Inc; 2001. pp. 96-9.
4. Blanca M, Vega JM, Garcia J, Carmona MJ, Terados S, Avila MJ, et al. Allergy to penicillin with good tolerance to other penicillins: study of the incidence in subjects allergic to beta-lactams. Clin Exp Allergy 1990;20:475-81.
5. Torres MJ, Romano A, Mayorga C, Moya MC, Guzman AE, Reche M, et al. Diagnostic evaluation of a large group of patients with immediate allergy to penicillins: the role of skin testing. Allergy 2001;56:850-6.
6. Blanca M, Romano A, Torres MJ, Férnandez J, Mayorga C, Rodriguez J, et al. Update on the evaluation of hypersensitivity reactions to betalactams. Allergy 2009;64:183-93.
7. Tsuji A, Nakashima E, Hamano S, Yamana T. Physicochemical properties of amphoteric β-lactam antibiotics, I: stability, solubility, and dissolution behavior of amino penicillins as a function of pH. J Pharm Sci 1978;67:1059-66.

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